Genome Browser:

“The Hitchhiker’s guide to the Genome”

User Manual

Copyright ©

All rights reserved. No part of this publication may be reproduced on any manner without the prior agreement and written permission of the publisher.

Version 1.1 - 2011/01/27

Table of Contents

1 Glossary 3

2 FAQ 4

3 General Information: 5

3.1 Architecture: 5

3.2 Main Features: 5

4 Basic Components 6

4.1 The Visualisation Field 7

4.2 My Projects 8

4.3 Tracks Menu 8

4.3.1 Static Tracks 8

4.3.2 Sample Tracks 9

4.4 GeneDigest 11

4.5 Project Tools 13

5 A project step by step 14

5.1 Open your project and tracks of interest. 14

1 Glossary

- Amplicons: An amplicon is a piece of DNA formed as the product of natural or artificial amplification events.
- BlazeDS: BlazeDS is the server-based Java remoting and web messaging technology of Adobe Open Source.
- MySQL: MySQL is an open source relational databasemanagementsystem (RDBMS), which uses SQL.
- Java: Java is a programming language.
- Lazy Data Fetching: Lazy Data Fetching is a design pattern commonly used in computer programming to defer initialization of an object until the point at which it is needed.
- Methylation Specific Primers: Methylation Specific Primers are primers used for methylation specific PCR.
- Ontology: Ontology is a means of classification.
- Oracle: Oracle is a databasemanagementsystem.
- PHP: Hypertext Preprocessor is a widely used, general-purpose scripting language that was originally designed for web development to produce dynamic web pages.
- Postgress: Postgress is an object-relational database management system (ORDBMS)
- Reference Genome: A reference genome is a digital nucleic acid sequence database, assembled by scientists as a representative example of a species' genetic code.
- Semantic Zooming: objects can render themselves differently when viewed at different sizes.

2 FAQ

1. Why aren't there any tracks in my project?

   You have to drag tracks (static and sample tracks) from the right bar into the Visualisation Field of the Genome Browser.

2. In Gene digest, how do we export all the methylated regions, and not just promotors, introns or exons?

   Uncheck the genes from this genelist box and make sure the checkboxes for the promotors, exons and introns are also unchecked. Because of the size of this data file, it may take a while to generate and download. Please try again if it should fail.

3. Why can’t I see the primers I designed?

   Primers are linked to a project, so most likely the problem will be solved by going to the project in which you designed primers. If this is not the case, click on the task list bottom-right and select show primers....

4. Where is the sample once I dragged it from the sample list on the right into the browser?

   This is a bug which we are working on. Reload the project if you need that sample again.

5. Why don't I see any transcripts in the transcript track?

   You have to select the gene of interest by clicking on it.

If you have any remaining and unanswered questions after reading this user manual, please contact Geert Trooskens by sending an email to the following email address: geert.trooskens@ugent.be and we will try to answer your questions as quickly as possible.

3 General Information:

3.1 Architecture:

The Genome Browser is a web-based application, meaning there is no need to install the software on your computer. The GB is system independent.

The Data is stored in MySQL, Postgres or Oracle, via PHP or java (BlazeDS).

The software is based on the Adobe open source Flex technology. The data is rendered at the client's side.

3.2 Main Features:

The software enables you to zoom from the chromosome to the nucleotide level and back.

Semantic zooming and lazy data fetching: Data is loaded and displayed according to the zooming level and region displayed.

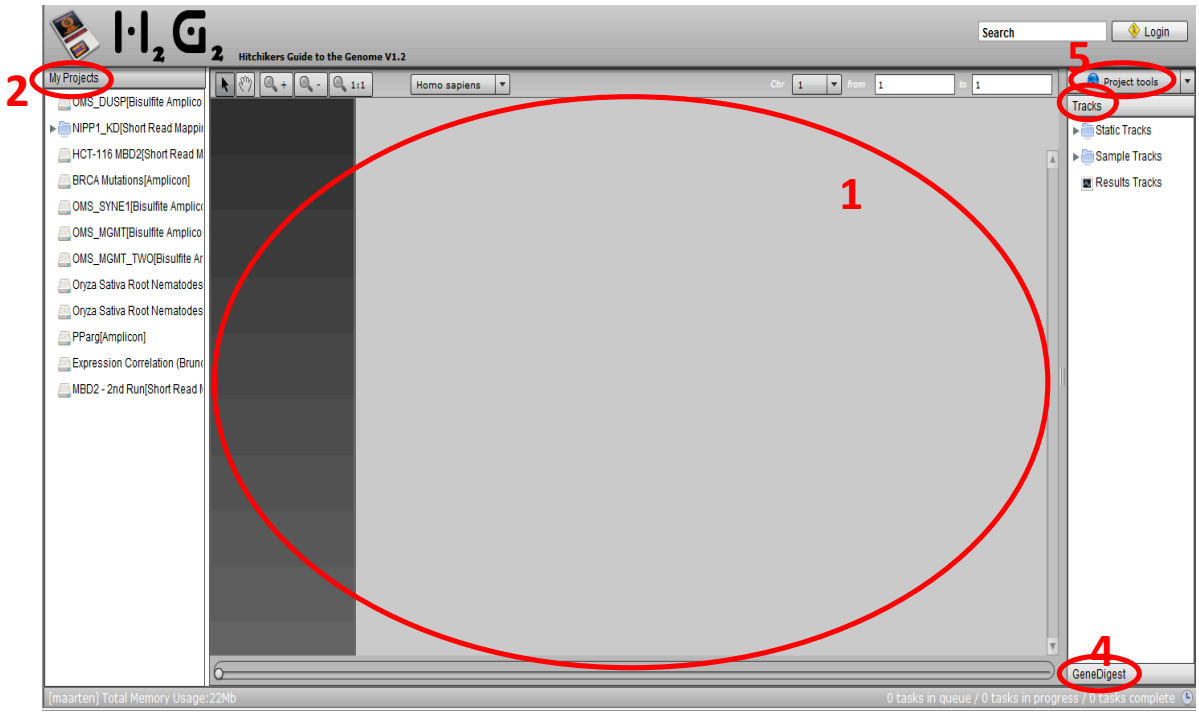
Interactive dragging and selecting.

Modular build, add tracks by adding data to the database.

4 Basic Components

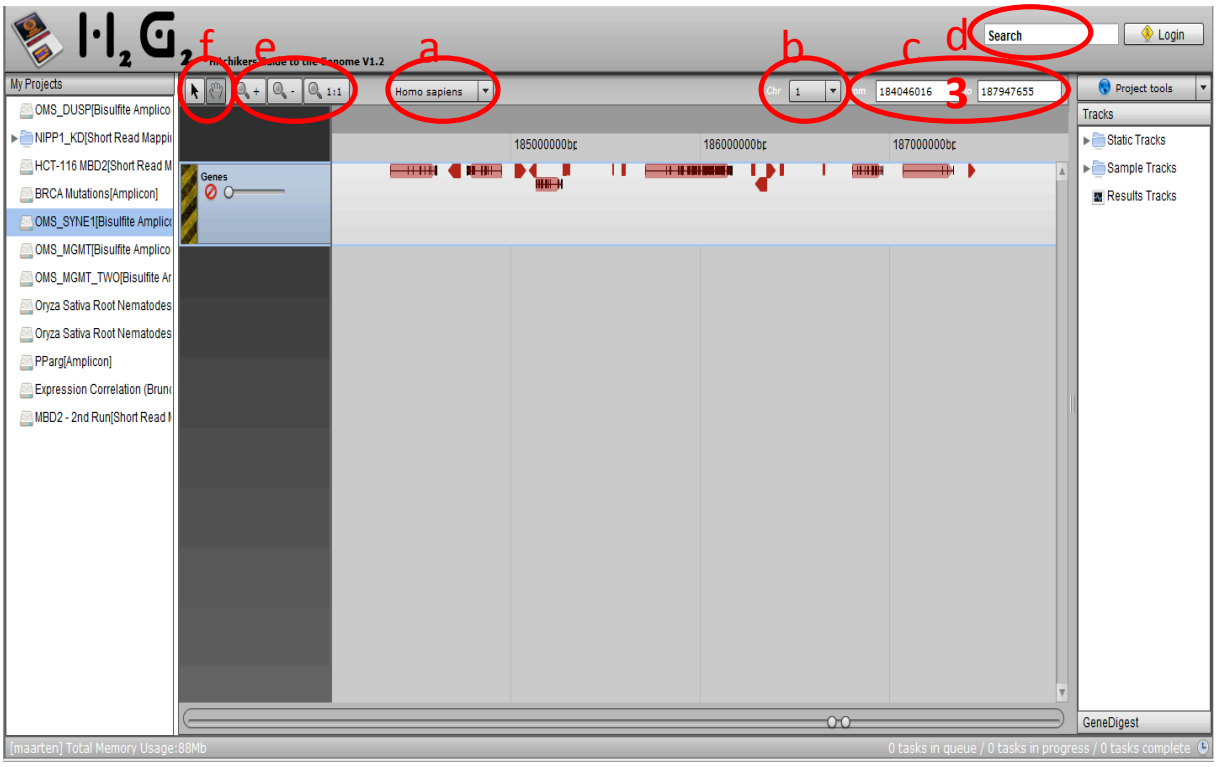
The Genome Browser software consists of 5 main fields.

1) The Visualisation Field
2) My Projects
3) Tracks Menu
4) GeneDigest
5) Project Tools



4.1 The Visualisation Field

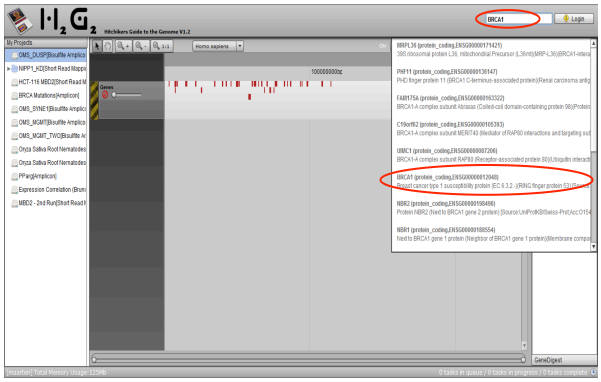
"The Visualisation Field" is the main field in the Genome Browser and is used to visualize and analyze experiment data.

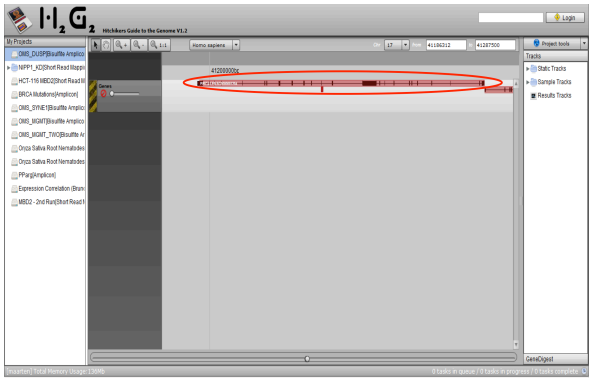


Several buttons in the visualization field enable the user the visualize the data of choice.

a. The user can select which reference genome needs to be shown in the visualization field.
b. The chromosome of interest can be selected for visualization.
c. On the chromosome of choice the user can specify which DNA region should be shown.
d. The search tool allows the user to enter the name of a gene of choice on which to zoom into.

   Enter a name in the search box at the right top corner of the Visualisation Field. A box will appear with all genes matching your search. Select the gene of choice and the Visualisation Field will automatically zoom into this gene.





e. Several buttons help the user to zoom to the level he or she desires. Secondly zooming can be performed by selecting a region in the visualization field itself. Simply click and drag over the region one wishes to show.

f. The user can browse through the genome/chromosome of choice by selecting the button with the figure of a hand on it. Then simply click in the visualization field, hold and move the cursor to the desired direction.

4.2 My Projects

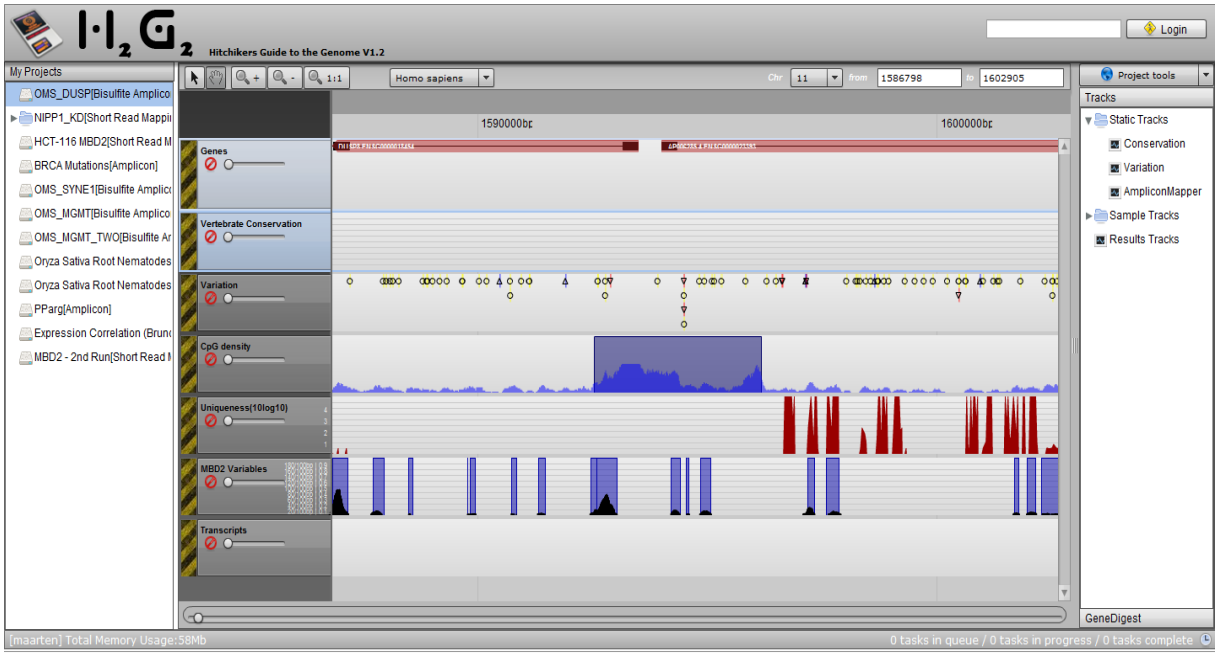
The Menu "My Projects" shows all the project/experiments the user has access to. From this menu one or more projects can be selected to be visualized on the visualization field (see 1).

To select your project, simply click on the project of choice in the list. Immediately the reference genome for your project (human, animal, plant, fungi, bacteria) will be shown in the visualization field and known genes will be visualized through red blocks in the "Genes Track".

4.3 Tracks Menu

4.3.1 Static Tracks

Static Tracks contain information that is independent from your experiment results. The information contained in the Static Tracks is information available to the general public, linked to the reference genome shown in the Visualisation Field.



- Conservation Track

  The Conservation Track shows which regions are conserved in other vertebrate species. The higher the peaks, the more this region is conserved in other species.

- Variation Track

  The Variation Track shows all known mutations in DBSNP .

- Amplicon Mapper Track

  When amplicons were designed and used in your experiment, the Amplicon Mapper Track will show the exact locations of these primers and amplicons.

- CpG Density Track

  The CpG Density Track shows the locations and density of CpGs in the DNA region.

- Uniqueness Track

  This track shows whether certain sequences can be found in other regions of the genome. Peaks show which regions can be found elsewhere in the genome.

- MBD2 Variables Track

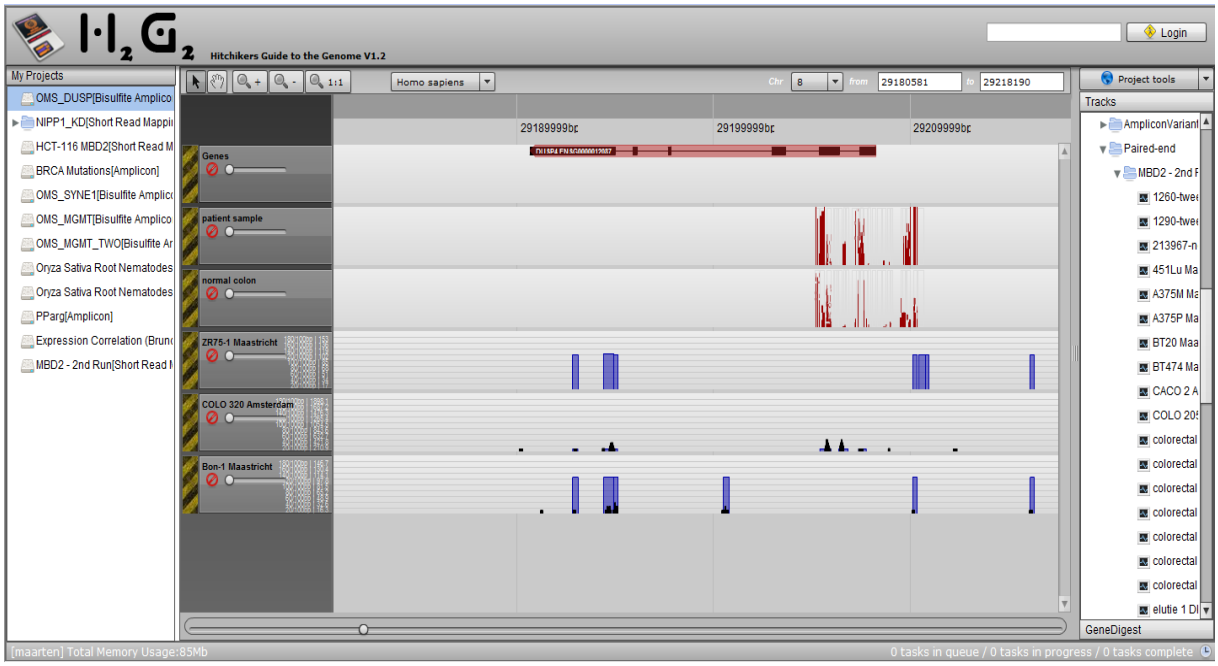
  The MBD2 Variables Track gives an overview of all methylated regions ever discovered/found by MBD2 sequencing experiments conducted at the University of Ghent/Biobix.

- Transcripts

  This track shows all known transcripts for the genes shown in the Genes Track.

4.3.2 Sample Tracks

Sample Tracks contain information that is linked to your experiment results. Only tracks linked to your experiment should be added to the Visualisation Field. Not all Sample Tracks will be discussed in this User Manual. Only Tracks important for epigenetics experiments will be explained.



- Waterfall Plot Tracks

  These Tracks show the results from Bisulfite Sequencing experiments through red peaks. Every peak is a CG location and the height of every peak shows the percentage of methylation.

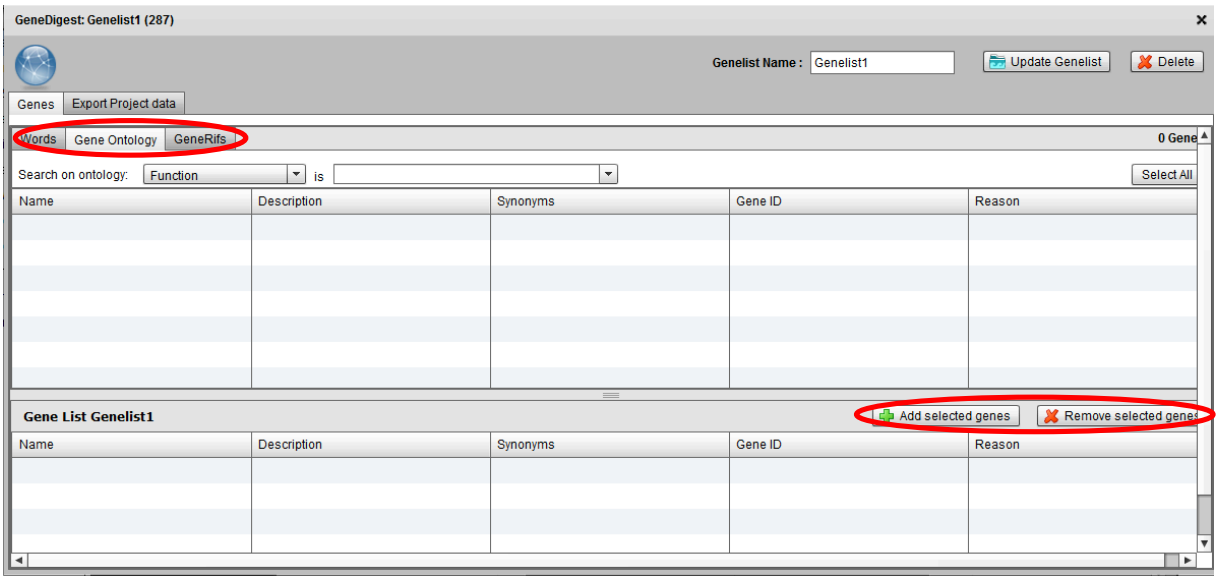
- Paired End Tracks

  These Tracks show the results from MBD2 sequencing experiments through black peaks. The height of the black peaks shows the methylation level in that specific region. Also purple blocks can be shown in these tracks. These blocks show the same data as the MBD2 Variables Track (see Static Tracks).

4.4 GeneDigest

The GeneDigest Tool allows users to make a list of genes and to export an exel file containing methylation info on all selected genes in all selected samples (selected earlier in the Visualisation Field). This exel file allows researchers to compare methylation levels in several genes of interest in samples of interest.

Click on GeneDigest and enter a name for your GeneList to enter the GeneDigest menu.



Inside the menu the user can search for genes either by:

- Words

Enter a gene name in the Words box.

- Gene Ontology

Genes will be searched by either function, process or component (the cell location where they come to expression (f.e. nucleus).

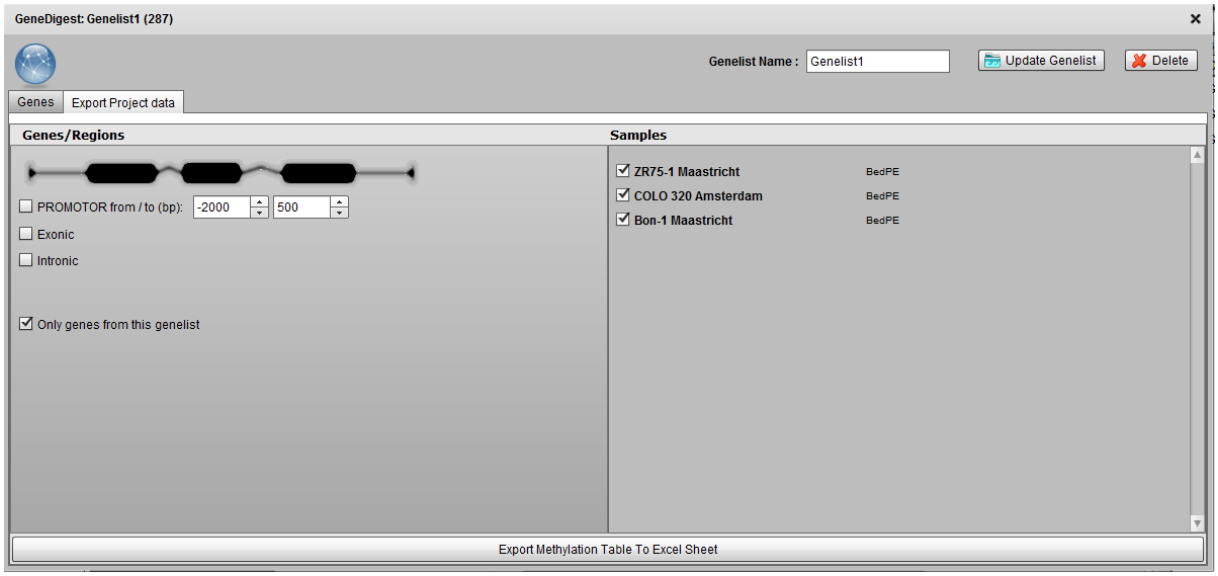
- GeneRifs

Enter text in the search box and your entry will be matched with published articles and the genes they refer to.

Finally add genes of interest to the genelist!

When the genelist has been constructed go to the “Export Project Data Menu”. Here the user can select which regions in which samples to export to the Exel file. When you uncheck the “Only genes from this genelist” box the exel file will be made for all known genes.

To export the data press the “Export Methylation Table to Excel Sheet” button.


GeneDigest: Genelist1 (287)
Genelist Name :
Genelist1
Update Genelist
Delete
Genes
Export Project data
Genes/Regions
PROMOTOR from / to (bp):
-2000
500
Exonic
Intronic
Only genes from this genelist
Samples
ZR75-1 Maastricht
BedPE
COLO 320 Amsterdam
BedPE
Bon-1 Maastricht
BedPE
Export Methylation Table To Excel Sheet

4.5 Project Tools

The Project Tools box contains 3 different actions.

- Print

The “Print” action allows the user to print the screen of the Genome Browser.

- Select Sequence

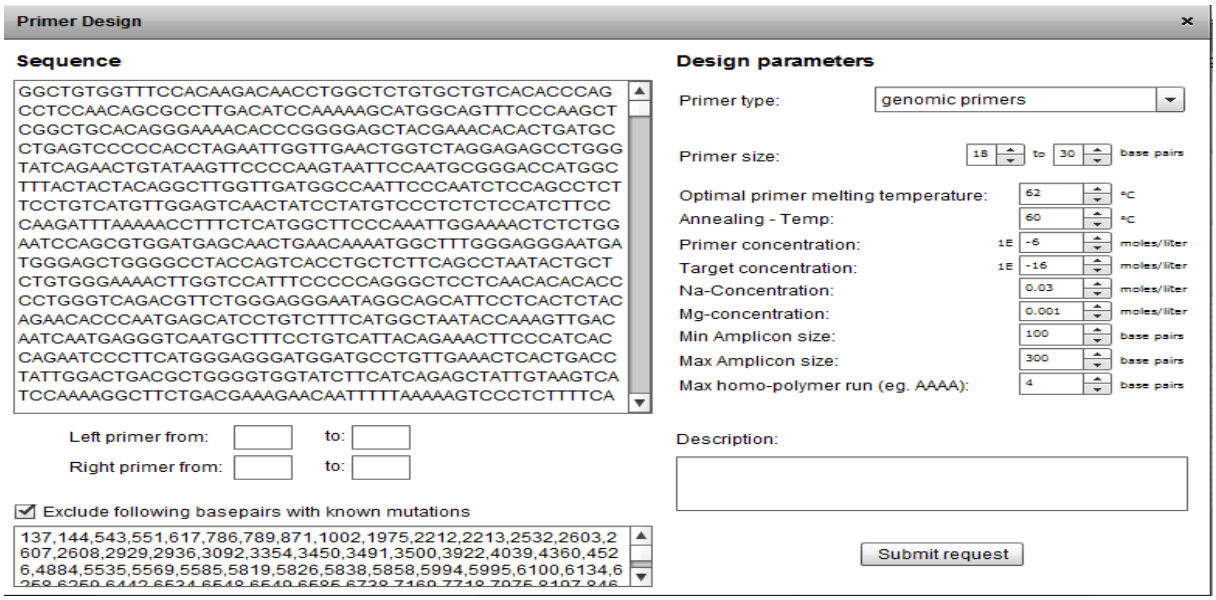
The “Select Sequence” action exports the DNA sequence shown in the Visualisation Field.

- Design Primer

The “Design Primer” action allows the user to design primers for future experiments. Several types of primers can be designed.

> First of all “Genomic Primers” can be designed. These primers can be used for amplicon sequencing experiments.
>
> Secondly “Methylation Specific Primers” can be designed. These primers can be used for methylation analysis experiments through PCR.
>
> Finally “Bisulfite Sequencing” primers can be designed. These primers are specifically designed for Bisulfite Sequencing experiments.

In the Primer Design box both the sequence of the region where the primer will be designed is shown together with the parameters impacting primer efficiency. The user can also chose to exclude known mutation sites from the primer design target region.

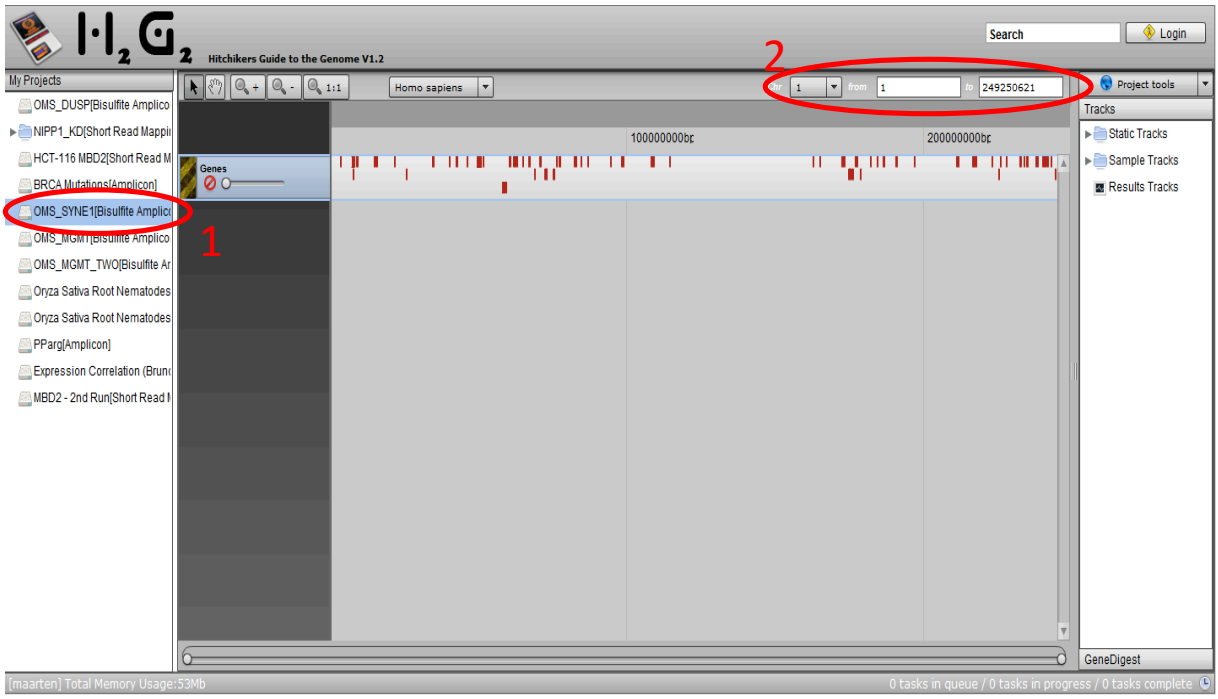


5 A project step by step

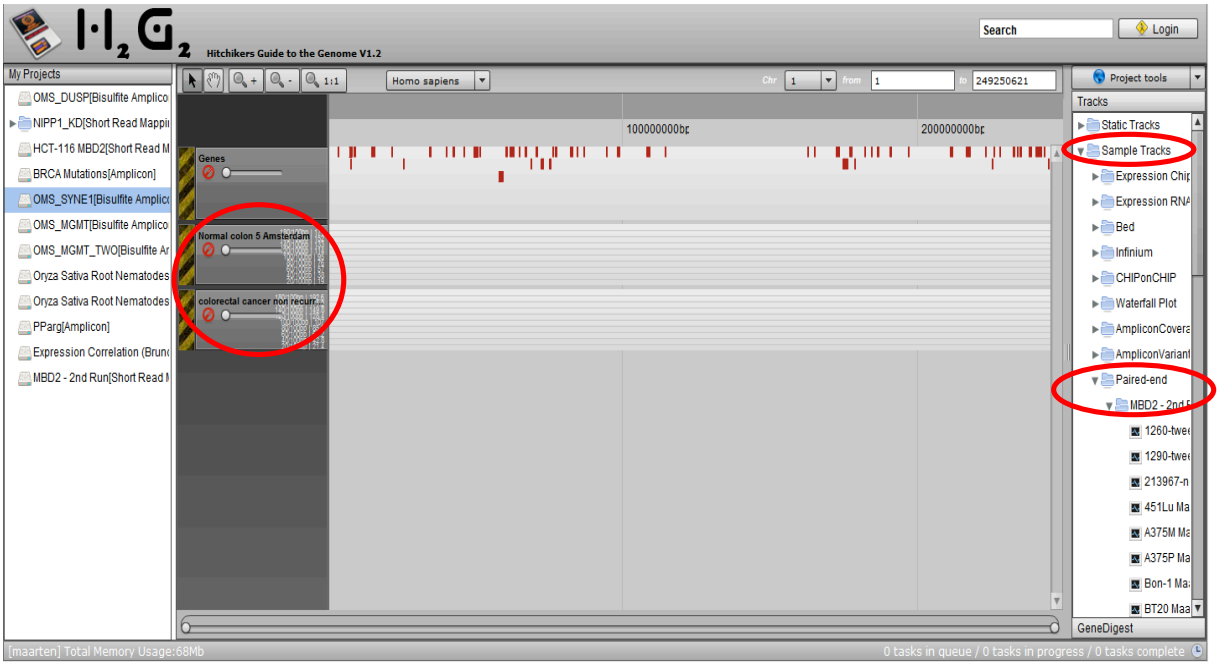
In this example we will take you through the different steps one should follow from opening the Genome Browser until interpreting your data and exporting them. In this case we will use the example of an epigenetics experiment, starting with MBD2 sequencing to find interesting differentially methylated regions.

5.1 Open your project and tracks of interest.

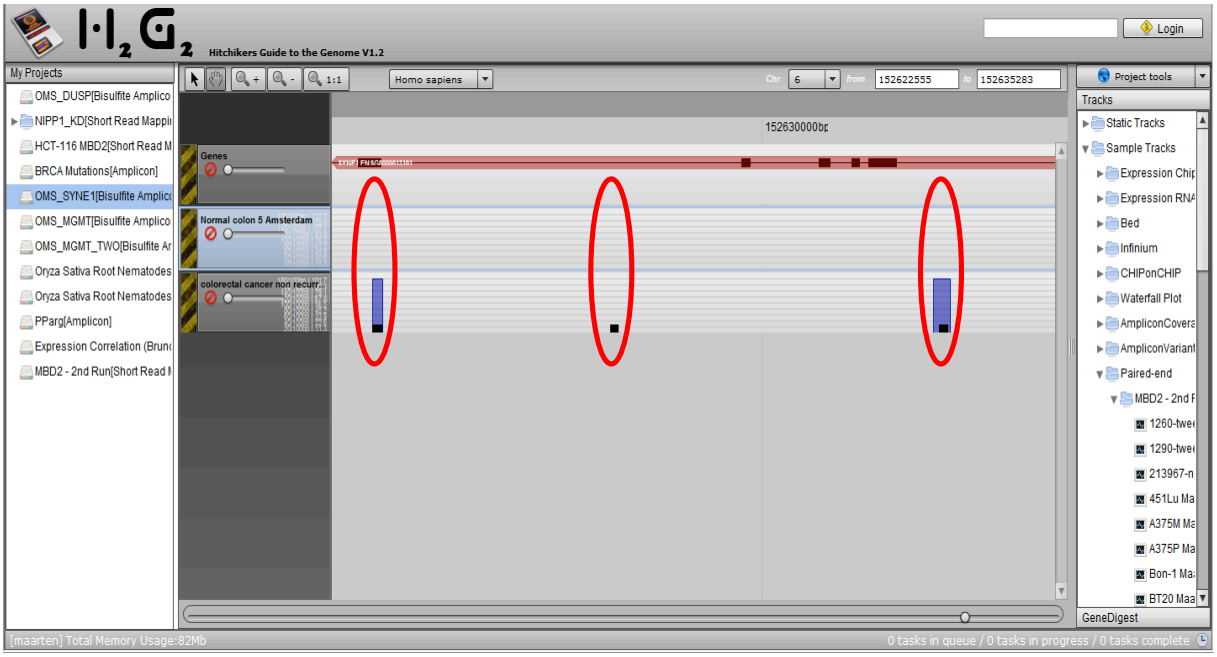
First of all open your project of choice(1). The genes track opens itself in the Visualisation Field. The user can see what region is shown at the upper right corner of the Visualisation Field(2). In the case shown below the first chromosome is visualized.



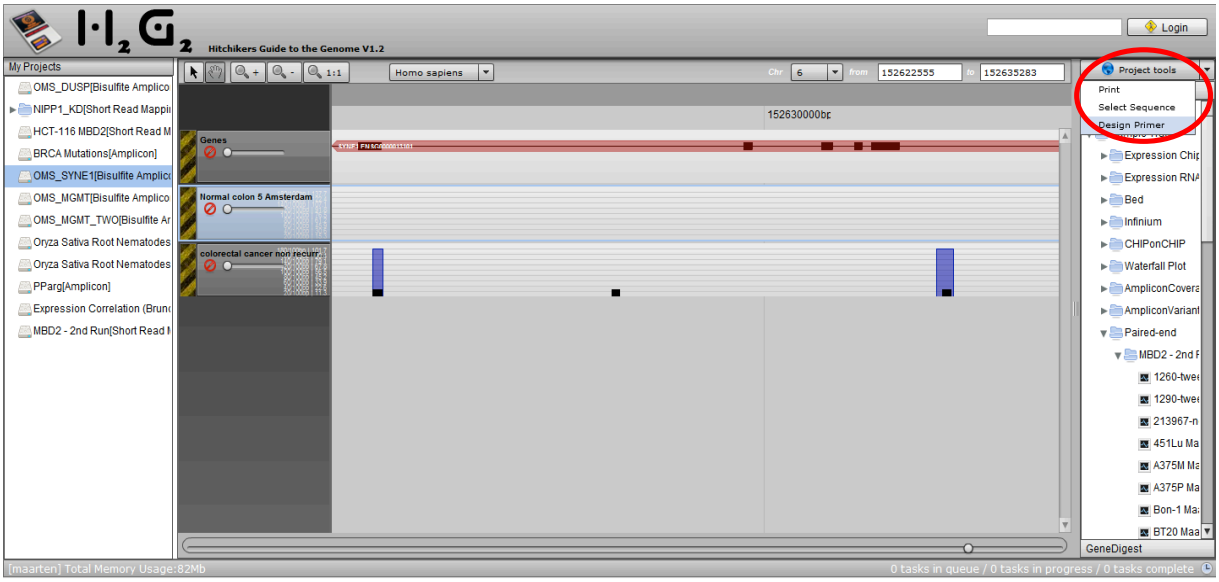
In our case an MBD2 sequencing run has been performed. Therefore the track that should be opened can be found under Sample Tracks – Paired End – MBD2 – 2nd RUN. Here all samples tested during the sequencing run can be found. In our example 2 samples will be visualized.



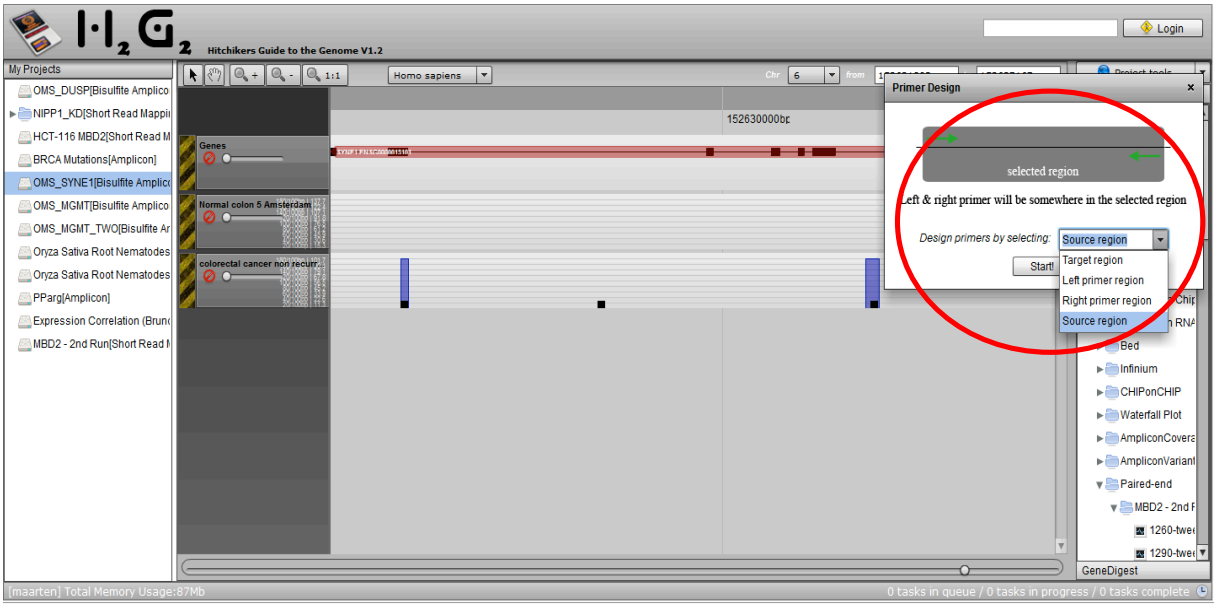
In first instance no peaks can be found in the two sample tracks. However after zooming peaks start to appear. Now the user can start to analyze his/her results. In our example differentially methylated regions are found between samples, meaning that this region can be interesting for more in-depth research through Bisulfite Sequencing.



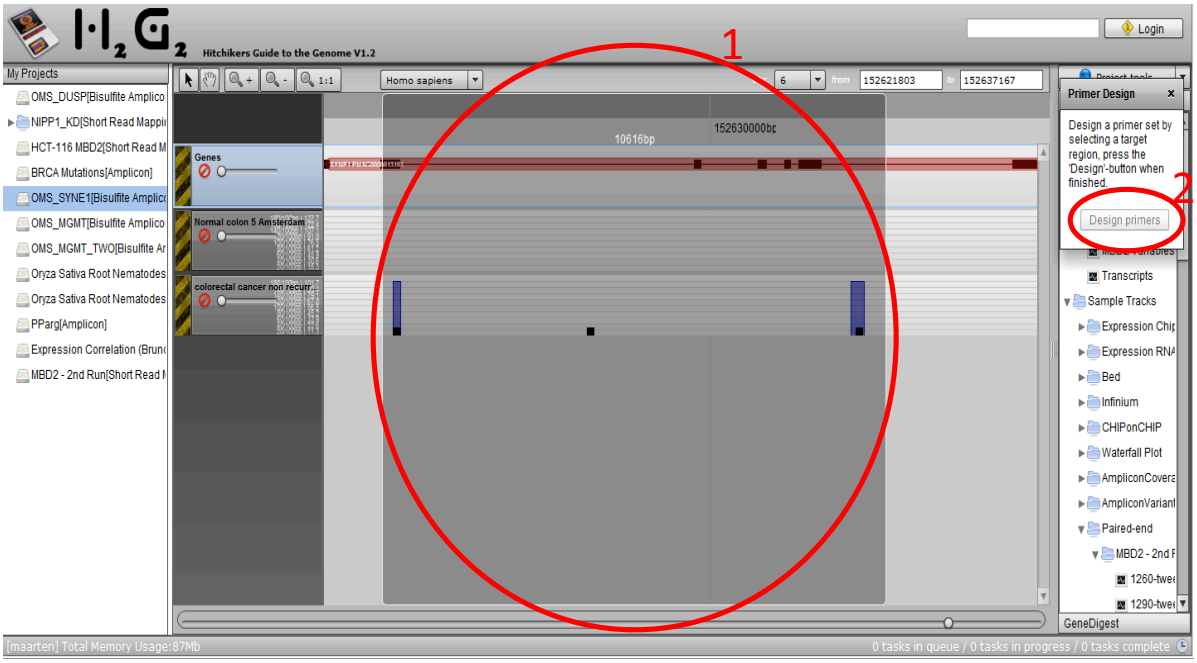
In case the user decides to continue his research by investigating the region more closely through Bisulfite Sequencing, the user will need to design primers specifically for his region of interest. The user can use the primer design tool provided in the Genome Browser. Simply select Project Tools – Design Primer.



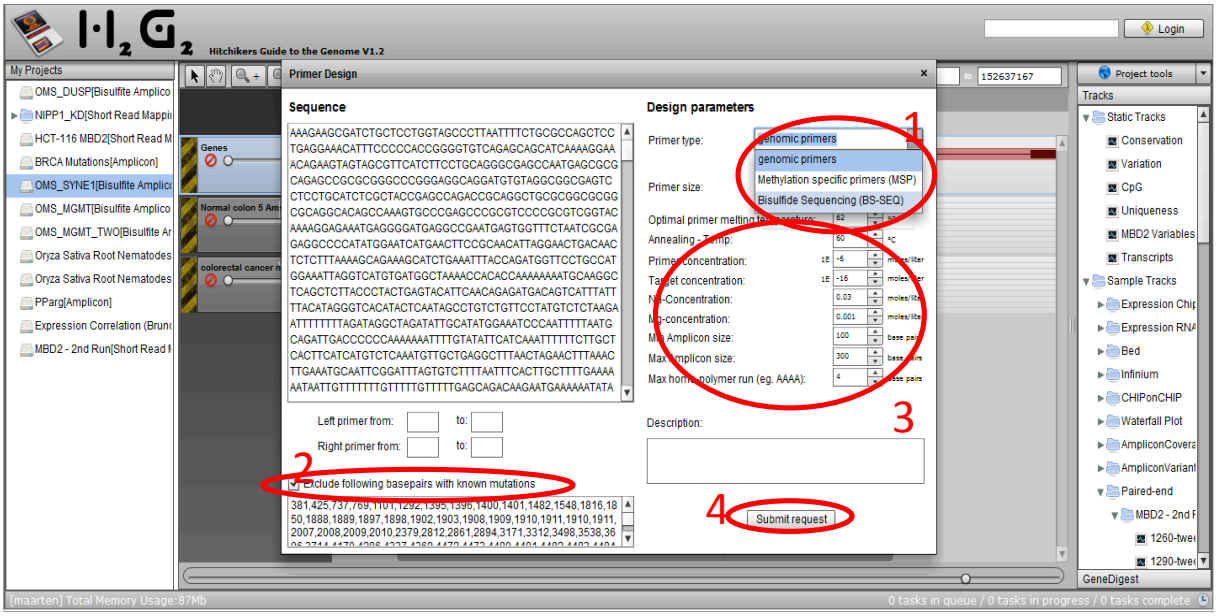
A drop down menu appears allowing the user to chose between different ways of designing the primers. The user can chose between either selecting the target region, the left primer region, the right primer region or the source region.



As soon as a selection has been made the Genome Browser asks the user to select the region in the Visualisation Field. Select the region of choice (1) and press the Design Primers Button (2).

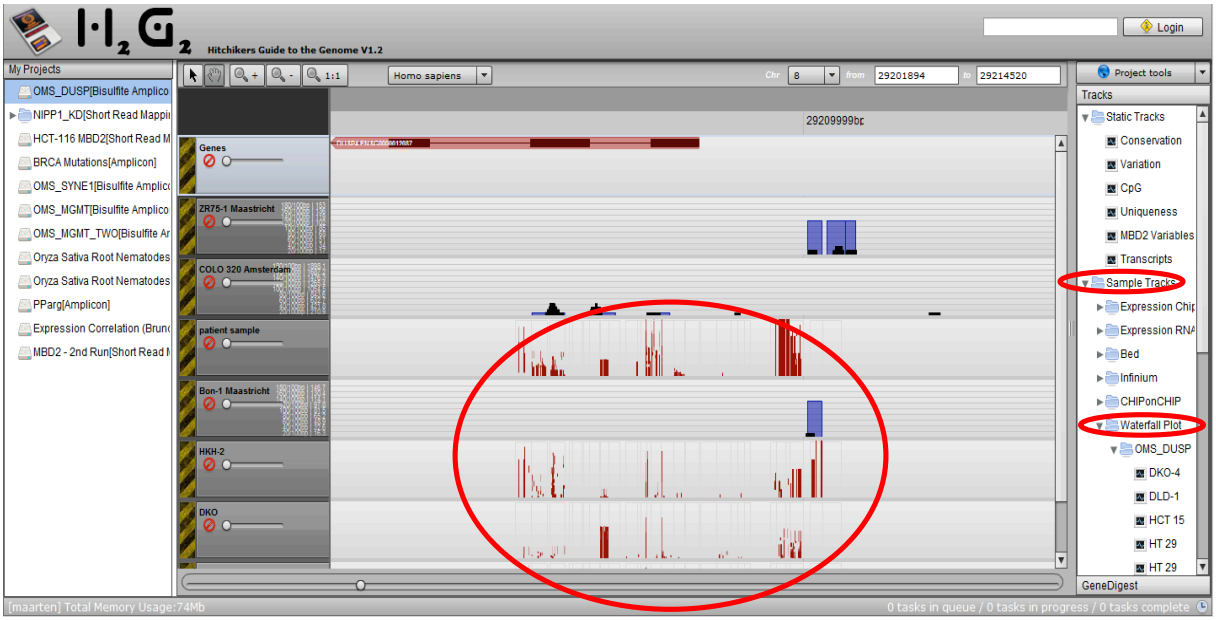


A new menu opens up and allows the user to select the type of primers he/she wishes to design (genomic primers, methylation-specific primers, bisulfite sequencing) (1). The user can chose whether or not to exclude known mutation locations from the primer design target region (2). The user can determine a set of parameters according to which the primers should be designed (3). When the user has finished his design specifications, he/she can submit his/her request.

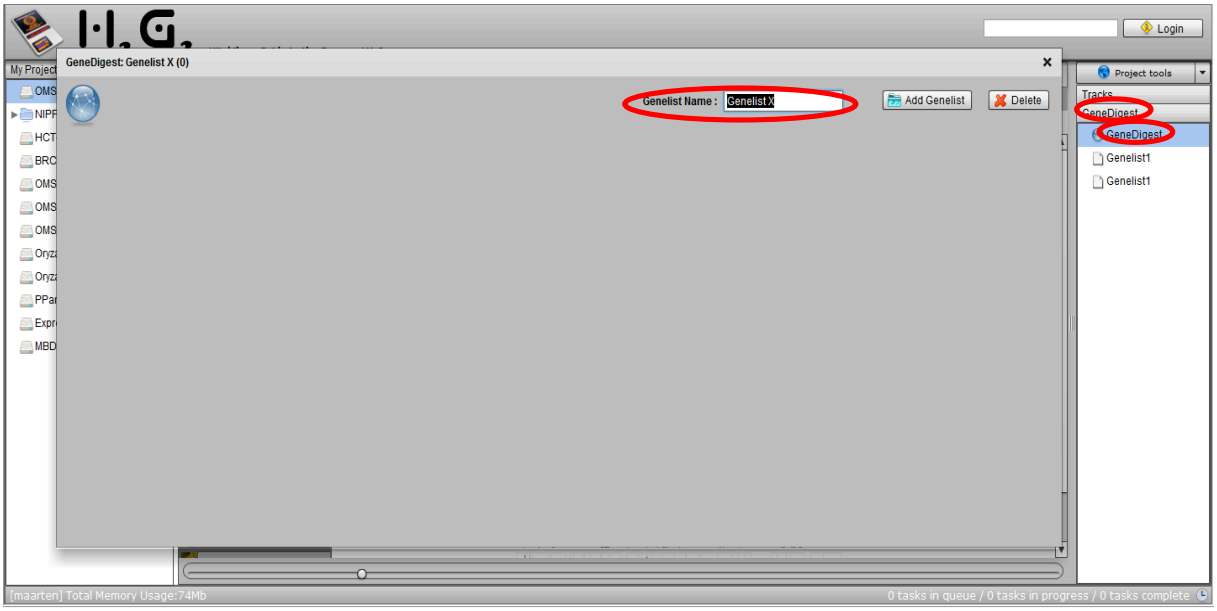


When the primers are designed and ordered, a Bisulfite Sequencing experiment can be executed. The user will be given access to these results as soon as the experiment has been completed. To open the results from the experiment in the Genome Browser, he/she should open Sample Tracks – Waterfall Plot – Project of Choice.

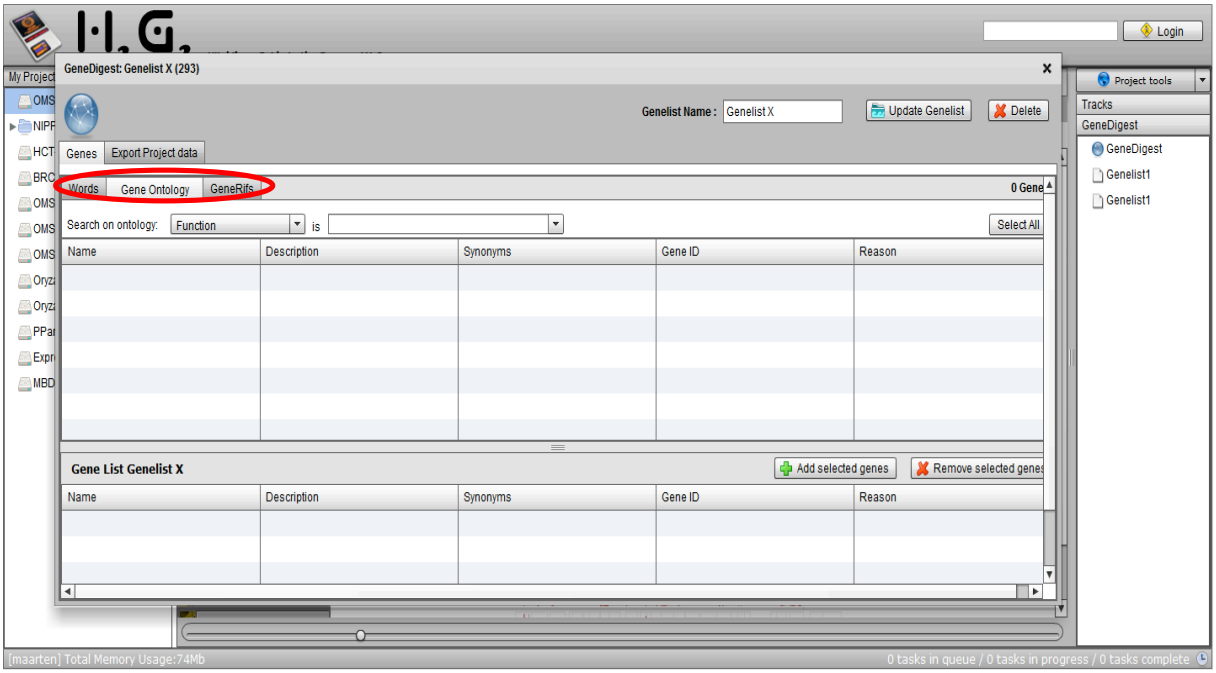
The user can add the tracks he/she wants to investigate to the Visualisation Field. These tracks show methylation on basepair level.



Finally the user can chose to export interesting data into an excel file. Therefore he/she should open the GeneDigest menu and enter a Genelist Name of choice.



Chose a set of genes. An overview of the methylation of the selected genes in the selected samples (Visualisation Field) will be given in an Excel file format.


Login
GeneDigest: Genelist X (293)
Genelist Name :
Genelist X
Update Genelist
Delete
Genes
Export Project data
Words
Gene Ontology
GeneRifs
0 Gene
Search on ontology:
Function
is
Select All
Name
Description
Synonyms
Gene ID
Reason
Gene List Genelist X
Add selected genes
Remove selected genes
Name
Description
Synonyms
Gene ID
Reason
Project tools
Tracks
GeneDigest
GeneDigest
Genelist1
Genelist1
[maarten] Total Memory Usage:74Mb
0 tasks in queue / 0 tasks in progress / 0 tasks complete